APPENDIX A

CLEAN COPY OF AMENDED PARAGRAPHS

Figure 13. Transduction efficiencies of MDA cells with Ad-LacZ at MOI's between 5 and 100. Expression of ß-gal was determined by x-gal staining 48 h after transduction. All additional experiments were performed at 50 MOI yielding an overall transduction rate > 80% with < 20% associated cytotoxicity.

In vivo radiosensitization in U-87 MG tumor xenografts after Ad-EGFR-CD533 infusion. To determine the effect of EGFR-CD533 on tumor radiosensitization, U-87 MG tumor xenografts measuring 8 to 10 mm in diameter were infused in vivo with AdLacZ or Ad-EGFRCD533 as described in "Material and Methods". This technique routinely yielded transduction efficiencies of 59 to 65 % (data not shown), as determined by x-gal staining of single cells, derived from tumor digests 3 days after AdLacZ infusion. Irradiation was performed three days after Ad infusion as described. In this study, 3 fractions of 3 Gy were used based on the in vitro studies showing enhanced radiosensitization with Ad-EGFR-CD533 transduction after repeated radiation exposures (Figure 33). Twenty-four h post-irradiation, tumors were digested to single cell suspension and ex vivo clonogenic survival was the treatment end point. The results presented in Figure 33 show that the treatment with Ad-EGFR-CD533 and radiation resulted in a 44% survival reduction relative to the control treatment with AdLacZ and radiation (10.4 vs. 18.5% survival; $P < 0.001$). The plating efficiencies of tumor cells from AdLacZ- and Ad-CMVEGFR-CD533- infused tumors were similar (6.79 vs. 6.14%, $p>0.5$).